Snake River Sockeye Salmon Captive Broodstock Program

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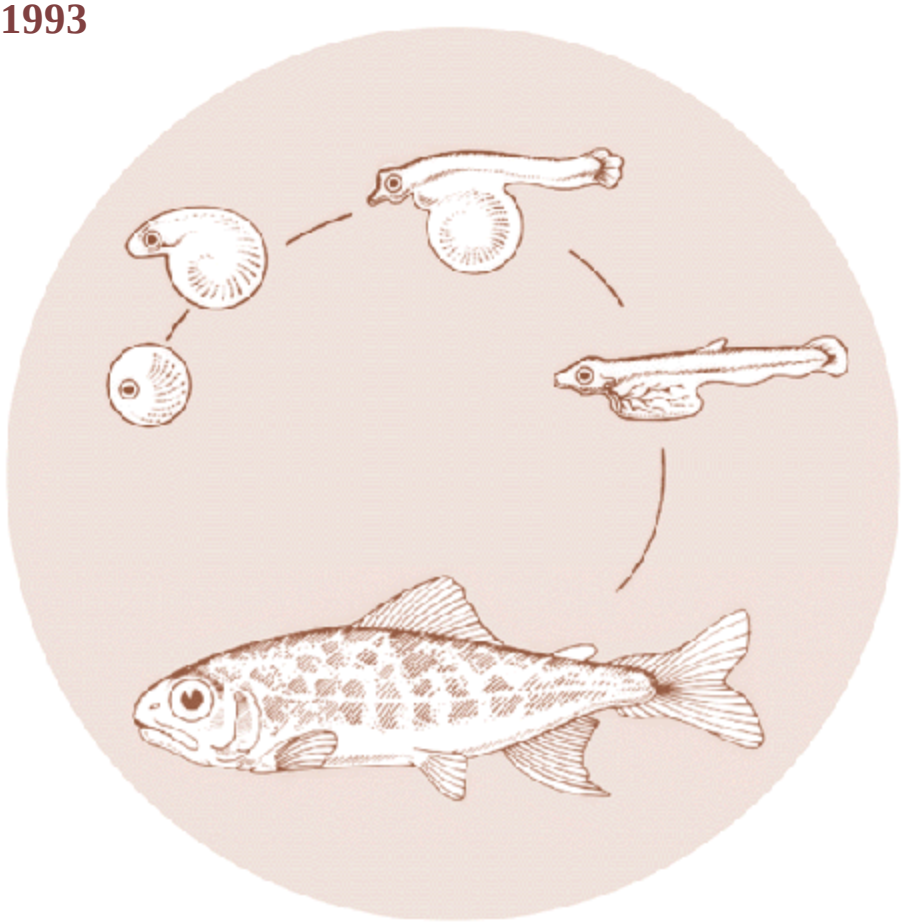

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SNAKE RIVER SOCKEYE SALMON CAPTIVE BROODSTOCK RESEARCH

ANNUAL REPORT 1993

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ABSTRACT

In 1991, the National Marine Fisheries Service listed Snake River sockeye salmon *Oncorhynchus nerka* as endangered under the Endangered Species Act of 1973. Initial steps to recover the species include the establishment of captive broodstocks at the Eagle Fish Hatchery in Eagle, Idaho. Research and recovery activities for sockeye conducted by the Idaho Department of Fish and Game during the period of April 1993 to April 1994 are covered by this report.

Eight anadromous adults (two female and six male) returned to the Redfish Lake Creek trap this year and were spawned at the Sawtooth Hatchery near Stanley, Idaho. Fecundity was 3160 for each female. The mean fertilization rate was 52% for female "A" and 65% for female "B." Captive broodstock also spawned as well as residual sockeye captured in a Merwin trap in Redfish Lake. Spawning data from 72 fish spawned during this period is included in this report.

Captive broodstock also matured later than normal (winter and spring 1994). Fish were spawned and samples were taken to investigate reasons for poor fertilization rates.

Twenty-four outmigrants of 1991 were selected for return to Redfish Lake for volitional spawning. Releases were made in August of 1993. All fish were implanted with sonic tags and tracking of this group began soon after the release to identify spawning-related activities.

A research project is being conducted on captive broodstock diets. The project will investigate the effect of diet modification on spawn timing, gamete quality, and fertilization rates. A second project used ultrasound to examine fish for sexual maturity. The goal was to obtain a group a fish to be released for volitional spawning. A total of 44 fish were found to be mature.

The performance of all captive groups held at Eagle are included in this report.

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INTRODUCTION

Historically, large numbers of adult sockeye salmon *Oncorhynchusnerka* were reported making the nearly 1,500 km journey from the ocean to the Stanley Basin lakes of central Idaho. Redfish Lake (Figure 1) is now the only lake in the basin which still supports runs of sockeye salmon (Bjornn et al. 1968). In 1991, as a result of only six fish returning to Redfish Lake during the previous three years, the National Marine Fisheries Service (NMFS) declared the Snake River sockeye salmon "endangered" under the Endangered Species Act (ESA) of 1973. The history of the Stanley Basin sockeye runs from 1982-l 993 is found in Table 1.

A technical committee has been established to coordinate efforts to recover this distinct population. The Stanley Basin Sockeye Technical Oversight Committee (SBSTOC) includes representatives of the Idaho Department of Fish and Game (IDFG), NMFS, Bonneville Power Administration (BPA), the Shoshone-Bannock Tribe (SBT), University of Idaho, and other state and federal agencies, as well as private groups involved in sockeye salmon restoration in Idaho.

In addition, NMFS is developing a plan to recover Snake River salmon (Snake River Salmon Recovery Plan 1993). This plan includes goals which should result in delisting the salmon. Currently, one draft goal is for at least 1,000 naturally-produced adults to return to Redfish Lake and at least 500 naturally-produced adults to return to each of two other lakes in the Stanley Basin.

Initial steps by IDFG to recover the species include the establishment of captive broodstocks at the Eagle Fish Hatchery (EAG) in Eagle, Idaho. Seven distinct broodstocks have been established consisting of progeny from Redfish Lake outmigrating smolts, anadromous adults, and residual sockeye. The current report summarizes activities from April 1993 through April 1994 by IDFG at EAG.

EAGLE HATCHERY

The Eagle Fish Hatchery was recommissioned in 1991 for rearing ESA-listed Snake River sockeye salmon captive broodstocks and their resulting progeny. The site is shared with the Eagle Fish Health Laboratory. Seven broodstocks have been developed for recovery from smolts, anadromous adults, and residual stocks.

Seven artesian wells are available for fish culture, and the water supply will be renovated in 1994 to add pumping capability and an intertie between two rearing areas. Effective volume available is approximately 0.2 m^3 per second. All culture is with first-use water in semi-square or round fiberglass tanks. Table 2 lists the inventory of tanks available for culture use.

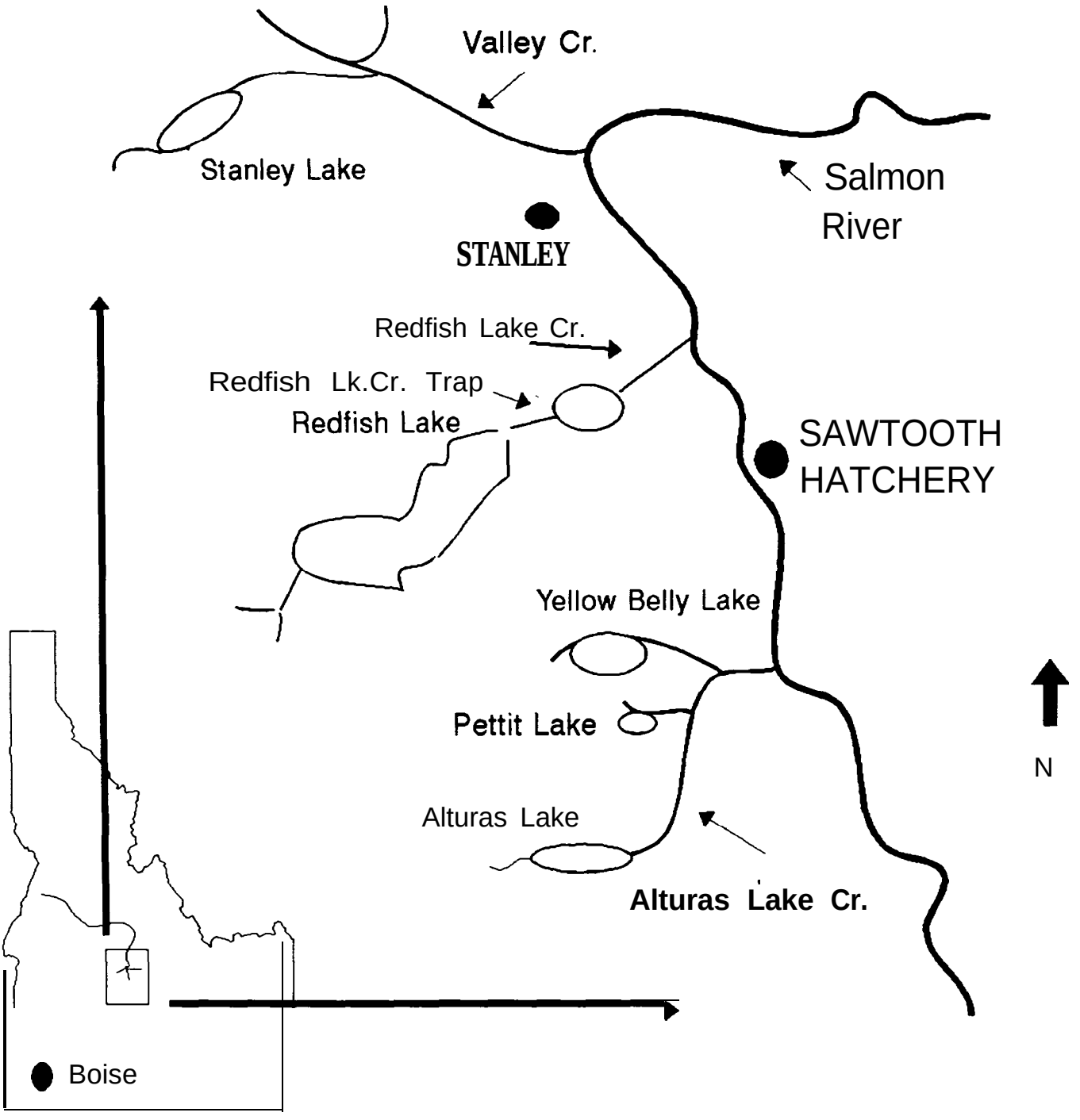

Figure 1. Stanley Basin Map.

Table 1. Recent run sizes of sockeye salmon passing Lower Granite Dam.

Date	Number of fish
1982	211
1983	122
1984	49
1985	35
1986	15
1987	29
1988	23
1989	2
1990	1[a]
1991	4
1992	1
1993	8

[a] Seen only at Ice Harbor Dam, Washington.

Table 2. List of existing facilities for incubating and rearing fish at the Eagle Fish Hatchery.

Hatchery rearing unit[a]	Unit volume (m^3)	Number of units	Construction
Incubator	NA	1,370	Plastic
R & D	0.09	48	Fiberglass
1 meter	0.30	52	Fiberglass
2 meter	1.42	34	Fiberglass
3 meter	6.50	38	Fiberglass
4 meter	8.89	4	Fiberglass

[a] Multiple, small rearing units are used to satisfy program genetic goals.

Rearing capacity for broodstock is limited by the capacity of a water chiller (– 6OO adults per year). Production capacity has yet to be determined, but should be approximately 150,000 presmolts per year with two size/time classes to be released into Redfish Lake or other lakes in the Stanley Basin.

PURPOSE

The purpose of the Eagle Fish Hatchery is to produce genetically-defined presmolts for restoration of Snake River sockeye salmon using captive broodstock technology. This program is designed to run for a single generation to limit the potential effects of long-term culture on the fitness of parental stock and progeny.

GOALS

Program goals are defined by the carrying capacity of receiving lakes within the critical habitat and by the genetic nature of indigenous 0. *nerka* populations. At this time, progeny of captive brood fish have only been planted into Redfish Lake of the Stanley Basin. In the future, presmolts will be released directly into the lake or released from net pens following a period of in-lake rearing at Redfish Lake.

OBJECTIVES

1. Maintain broodstocks separately with identification of the genetic origin of individuals to maximize diversity and minimize inbreeding during spawning.

2. Define best management plans for Eagle Hatchery to be used for recovery of listed species.

3. Produce defined groups of progeny for evaluation of release strategies, appropriate size/time targets, and genetic origin to maximize smolt performance and adult return.

4. Apply principles from the Alaska Sockeye Salmon Culture Manual (1994) to reduce the risk of infectious disease interfering with attainment of program goals.

CURRENT PRACTICES TO ACHIEVE THESE OBJECTIVES

Hatchery Production

1. All broodstocks originating from anadromous adults are duplicated at the NMFS, Big Beef Creek (BBC) facility in Washington State, in the case of a total facility disaster. Progeny from the BBC program are returned to EAG only as iodophor-disinfected eyed eggs

2. Maintain rearing densities at or below 8 kg/m^3 at least through smolt size.

3. Fin-clip all production fish released to Stanley Basin lakes for identification purposes upon return as adults. Appropriate numbers of progeny are PIT-tagged for evaluation of smolt performance at migration.

4. All rearing units are kept in quarantine to prevent disease transfer.

5. Currently, up to 20 adults, which return to the weir on Redfish Lake Creek, are to be used for hatchery broodstock. All adults in excess of 20 are to be released upstream for natural spawning.

Minimize interactions with other fish populations through proper rearing and release strategies

All returning anadromous adults have been taken into captivity since 1991. As a result, the only progeny in Redfish Lake with direct anadromous origin are the result of enhancement. There is a small population of "residual" sockeye in Redfish Lake which continues to reproduce annually. Release strategies for progeny are designed to maximize migration of smolts and minimize residualization. Control efforts targeting the existing kokanee population are to reduce competition with *0. nerka* possessing anadromous capability. Research into the best release strategy is on-going and deals with size/time and genetic origin of the presmolts. The goal is to allow for cultured fish to reside in Redfish Lake over winter and migrate volitionally in the spring.

Maintain stock integrity and genetic diversity

National Marine Fisheries Service Propagation Permit #795 requires that IDFG consult with NMFS annually to establish a spawning matrix for those groups which are expected to mature. The goal of this consultation is to evaluate how groups can be spawned to maximize diversity and minimize inbreeding with the limited genetic base available in this program. Each brood fish held in this program has been PIT-tagged and can be traced back to the origin of both parents. This is used to direct mate selection and reduce the probability of inbreeding.

Management of Bacterial Kidney Disease in broodstocks

Bacterial kidney disease (BKD) has been observed as a cause of death at EAG in a single group of fish (OM91, group 4) (see Table 3 for broodstock definitions), and this group was transferred to Sawtooth Fish Hatchery (SFH) in 1993. BKD has been observed at BBC in BY91, BY93, and OMBY9IBY93. The spawning of BY91 at BBC and OM91 Group 4 at SFH in 1994 creates a risk of importing BKD to EAG with the eggs originating from these broodstocks. Idaho Department of Fish and Game proposed that all parents be screened for BKD using the Enzyme-linked-immunosorbent-assay (ELISA) procedure and that only those eggs originating from parents with ELISA optical density (OD) values <0.2 be imported to EAG. Eggs from parents with ELISA OD values between 0.2 and 0.4 are to be reared at the station of origin and eggs from parents with ELISA OD values >0.4 are to be destroyed.

PERFORMANCE OF CULTURED GROUPS

OM91

The Redfish Lake outmigrants retained for broodstock in 1991 started this report period with an inventory of 703 fish and ended with 375 for 53% survival. Fish weight increased from 335 g to 1,800 g. Bacterial kidney disease was detected in group 4 in 1991. Erythromycin treatments were applied to group 4 and showed efficacy (NPDES permit # ID - 002274-8). The other groups have remained free of detectable BKD using ELISA and Fluorescent Antibody Test (FAT) diagnostic techniques. Strict quarantine practices are applied to these groups. See Table 4 and Appendix D for mortality and growth information.

Several significant events occurred during the April 1993 to April 1994 period in the culture of the OM91 groups. The first event was the discovery of tumors, generally rare in salmonids. At least nine individuals of groups 1, 2, 3, and 5 developed tumors. The most prevalent was a thyoma which appears as an enlargement of the thymus. The growth attained the size of a golf ball in fish weighing approximately 900 g. This has been previously reported in salmonids and classified as a lymphosarcoma (Smith, 1971; Smith, 1993). Histological sections demonstrated that the cell type was a lymphocyte and the tumor was sent for confirmation to the Institute For Tumors Of Lower Vertebrates. Attempts to culture the cells of this tumor are in process.

A second type of tumor was collected from the isthmus of one fish during the same winter. This tumor was identified as a fibroma and has also been reported previously in salmonids. These tissues were also submitted to the lnstitue For Tumors Of Lower Vertebrates for verification. There has been some speculation that susceptibility to tumors has a genetic base in salmonids. This may be evidence that this population has undergone a single or series of genetic bottlenecks previously. If that were true, there also should be a high incidence of tumors in the progeny of these broodstocks. It may be difficult to examine this occurrence because the progeny will mostly be at sea if these tumors develop at the fourth year of life as they did in captivity.

Table 3. Abbreviations used in the text.

The following abbreviations are employed throughout this report for the sake of brevity. Abbreviations are used for accuracy in describing the origin of stocks used in this recovery effort and are listed in chronological order.

DESIGNATION	PARENTAL ORIGIN
OM91	**Obtained as outmigrants from Redfish Lake in April, May, and June of 1991. Cultured at Eagle Hatchery from May 1991 until the present and spawned in 1993.**
ALTOM91	**Obtained as outmigrants in May and June 1991 and cultured at Eagle and Sawtooth hatcheries until released into Alturas Lake in the fall, 1993.**
BY91	**Obtained as eggs by spawning a single female and three male anadromous sockeye which returned in 1991. These were divided between NMFS and IDFG as eggs and reared at Eagle, Montlake, and Big Beef Creek hatcheries. Some maturation occurred in 1993.**
OM92	**Outmigrants from Redfish Lake Creek obtained in 1992 and cultured at Eagle to the present.**
ALTOM92	**Outmigrants trapped in 1992 and raised at Eagle and Sawtooth hatcheries until released to Alturas Lake in the fall, 1993.**
RESBY92	**Progeny of three residual sockeye seined from Redfish Lake in November 1992 and reared at Eagle until the present.**
OM93	**Outmigrants obtained as smolts in 1993 and reared at Eagle to the present.**
BY93	**The progeny of the matings of two female and six male sockey which returned in 1993. The eggs were divided between Eagle and Big Beef Creek hatcheries. Those at Eagle were destined for broodstock and release, while those at BBC were for broodstock only.**
RESBY93	**Progeny of residual sockeye seined from Redfish Lake in November 1993 and reared at Eagle until the present.**

Table 4. Trapping and rearing mortalities for Redfish Lake sockeye, April 1991 to April 1994.

	Broodstock				
	OM 91	BY 91	OM 92	OM 93	RES 92
Starting Inventory	861	937	79	48	35
Trapping	58	0	0	0	0
Transport	22	0	0	0	0
Pinheads	0	0	3	0	0
Handling	53	7	0	0	0
Jump-outs	10	6	1	1	0
Bacterial Gill Disease	29	2	4	0	0
Other Infections	97	9	0	0	0
Tumors	9	3	0	0	0
Spawners	4	4	0	0	0
Prespawners	3	0	0	0	0
Dewatered	108	10	0	25	0
Sacrifice	12	7	0	0	0
Undetermined	81	42	2	0	1
Ending Inventory	375	847	69	22	34

Space and water constraints due to delays in the completion of facility expansion at EAG forced the move of OM91 groups 1&2 to SFH in April 1993. This was not an appropriate time for such a move because the water temperatures at the two stations are very different (EAG = 12.8°C; SFH = 4.5°C). The change in temperature caused the fish to go off feed for several weeks, and a *Pseudomonas sp.* infection was exacerbated by the stress. The loss over the next two months amounted to 37%. Once feeding was initiated by the use of a highly palatable feed formulated by Bioproducts (Warrenton, Oregon), Oxytetracycline was used to counter the infection. Mortality decreased as the fish adapted to the rising well water temperatures as the spring progressed.

Each mortality in the OM91 group was sampled for genetics and pathology. No BKD has been demonstrated in groups other than group 4; however, bacteria causing cold water disease, motile aeromonad septicemia, and bacterial gill disease have been the primary cause of death. Mortalities were also processed for gill raker counts, flesh tone due to the carotinoid added to the diet, number and distribution of dorsal spots, and condition of the gonads. Most of these data were collected to add to the differentiation between the anadromous and resident forms of *0. nerka.* These variables may offer a more rapid verification technique than protein gel electrophoresis or DNA based tests.

We have also observed an enlarging of the ovaries in about 60% of the females that died during the late winter and spring. Ova diameters were increased in the larger of the ovaries. This may be used as a predictor of impending sexual maturation in fall 1993.

ALTOM91

Alturas Lake outmigrants captured in spring 1991 were cultured at EAG until September 1992 when they were moved to SFH due to space and water constraints. The starting inventory was 136 and the ending inventory was 125. The overall survival has been 90% for the two years of culture and a weight gain of 888 g for the period. The causes of mortality have been jumping out of rearing tanks and motile aeromonad septicemia. No BKD has been detected in the mortalities; however, two prophylactic treatments with Erythromycin have been applied during the April 1992 to April 1993 period. The mortalities have been examined and sampled as described above for OM91, and the increase in ovary size has been noted for the females of this group as well. All remaining fish were released into Alturas Lake during August, September, and October of 1993 to spawn volitionally.

RFLBY91

These fish are the progeny of a single female and three males which returned to Redfish Lake as adult sockeye in 199 1. Progeny were cultured as separate subfamilies until they were PIT-tagged in June 1992. The subfamilies were from male A, B, and C; a group that was fertilized with pooled milt from all three males (ABC); and those eggs recovered from volitionally spawned eggs (NAT). These fish grew to 1,240 g by April 1994 with a loss of 90

fish (Appendix D). Fish were PIT-tagged at 7 g and randomized into groups to prevent loss of an entire genetic subfamily in the event of a single tank disaster. Mortalities for this group are presented in Table 4. This group is also represented by a similar number of fish being reared by NMFS at the Montlake, Washington facility.

The cause of death in the mortalities was varied and included cold water disease, motile aeromonad septicemia, bacterial gill disease, pinheads, and jump-outs. All mortalities were examined for BKD and viruses with negative results. Three tumors have been noted in these fish.

OM92

Seventy-nine outmigrants captured from Redfish Lake Creek in 1992 were taken into culture averaging 8.7 g. In April 1994, there were 69 remaining averaging 966 g. The causes of this loss were pinheading, bacterial gill disease, and one jump-out. No BKD was detected in this group. Growth was uniform, although a slowing of growth was noted in the first winter (Appendix D) This may have been a physiological effect because the well water temperature at EAG only decreases slightly in response to lower air temperature. Mortality data can be found in Table 4.

ALTOM92

Ninety-five outmigrants were taken into culture at EAG averaging 10 g in May 1992. In November 1993, the 75 remaining fish were released into Alturas Lake at an average size of 463 g. The cause of death during culture at EAG was determined for seven fish. The causes included two pinheads, two with surface *Pseudomonas* lesions, two with gill cysts resembling *Loma,* and one which died from handling during a monthly inventory. The growth was biphasic with a decrease in winter months as was seen with OM92.

RESBY92

Thirty-six fertilized eggs were obtained from the mating of a partially-spawned residual sockeye female and two mature males captured from Sockeye Beach of Redfish Lake by Dr. Ernie Brannon, University of Idaho. The number of CTU to eye was 318, to hatch was 614, and to swim-up and first feeding was 932. Yolk absorption was fatal to one fish, but all others have done well. The fecundity for the female could not be determined. Progeny will be cultured at EAG for use in the captive broodstock program and represent an ESA-listed component of Snake River sockeye. As of April 1994,34 fish weighing 175 g each remained. Growth for the group can be found in Appendix D, while mortality data can be found in Table 4.

RESBY93

The progeny of two females and four males from the residuals trapped in 1993 were divided between EAG and BBC. The inventory at EAG was 44 fish at 0.8 g each on March 1, 1994, and currently there are 30 fish at 1 12 g each. The primary cause of mortality for this group was jump-outs. Those fish remaining will be mated with RESBY92 to evaluate the utility of this stock for recovery. Maturation is expected in 1995. See the Adult Trapping section (this report) for details on mating crosses and number of fish captured in 1993.

OM93

Fourty-eight outmigrants were delivered as four groups to EAG, and feeding was initiated using the krill, blood worms, Biodiet, cat food, and anchovy paste formulation which was successful in 1991 and 1992 (Johnson 1992,1993). The fish averaged 8.9 g in weight and 98.5 mm fork length when delivered and had grown to 250 g by April 1994. A mistake during tank cleaning caused the loss of 25 fish in October 1993. Ending inventory on April 1994 was 22 fish. See Table 4 and Appendix D for mortality and growth information.

ADULT TRAPPING AT REDFISH LAKE CREEK

The adult trap on Redfish Lake Creek was used in 1993 as in the previous two years. The trap was installed on July 6 and operated through October 25, 1993. Six male and two female sockeye were trapped in August and transported to SFH. Fish were held in well water with gravel from Sockeye Beach of Redfish Lake as substrate. This technique was used successfully in 1991 and 1992. Idaho Department of Fish and Game personnel examined the adults twice weekly, and each fish received a PIT-tag. Cryopreserved sperm from the single male which returned in 1992 was brought to SFH. Spawning was done on October 8. Eggs were harvested and divided Into six equal lots with a seventh lot of 15 g used with the cryopreserved sperm. Motility was examined for each fresh sperm collection. Eggs of each cross were incubated as separate lots at SFH in well water.

Sperm samples from each anadromous male were harvested for cryopreservation. Samples were shipped by air and cryopreserved in duplicate by the laboratories of Dr. Joe Cloud (University of Idaho [UI]) and Dr. Gary Thorgaard (Washington State University). Fertilization was tested with kokanee eggs and selected cryopreserved milt samples from both groups.

Appropriate duplicate tissue samples for genetic identification and pathology were removed when each adult was sacrificed at spawning or after milt was no longer available. These were distributed to Dr. Waples (NMFS), Dr. Brannon (UI), and the Eagle Fish Health Laboratory (IDFG) for analysis. The carcasses were delivered to EAG, sampled, and incinerated.

TRAPPING OF RESIDUAL SOCKEYE IN REDFISH LAKE FOR 1993

A floating Merwin trap was installed October 18, 1993 in Redfish Lake to capture sexually maturing residual sockeye salmon for inclusion in the captive broodstock program. The trap (located near Sockeye Beach) remained in operation until October 29, 1993. Personnel from the IDFG Sawtooth Hatchery emptied the trap daily. Twenty-four *0. nerka* were captured in 1993. Non-targeted fish species captured in the trap were identified, enumerated and released unharmed. The results of trapping and destiny of trapped residual fish are listed in Tables 5 and 6.

SPAWNING ACTIVITIES

Brood Year 1993 (October and November 1993)

Anadromous Matings

The two female and six male anadromous sockeye were spawned on October 8, 1993 (Table 7). The protocol employed for these spawners was the same used in 1991 except that the cryopreserved sperm of the single male of 1992 was used on a small proportion (15 g) of the eggs of each female. Fertilized eggs were incubated as separate lots until eyed and divided between EAG and BBC for rearing. No fertilization was obtained with the cryopreserved sperm.

Fecundity was 3,160 for each female. Males #2 and #5 did not fertilize eggs very well compared with the others. The mean fertilization rate was 52% for female "A" and 65% for female "B."

Big Beef Creek was shipped 518 eggs from the crosses made with female "A," while 1,143 were incubated at EAG. BBC was shipped 675 eggs of female "B," and 1,359 were retained at EAG. The goal was to have eggs for broodstock at each facility, and fish in excess of broodstock capacity would be reared at EAG and returned to Redfish Lake as presmolts in 1994.

Matings of Captive Broodstock

Table 7 and Appendix A list the matings made from captive broodstock adults which matured during this period. The spawning matrix utilized anadromous males when possible (14 OM91 females). Two males of OM91 and three BY91 males were also used with these females. A single BY91 female matured and her eggs were fertilized with two BY91 males.

Table 5. Merwin trap results for residual sockeye at Sockeye Beach, Redfish Lake, October 19-29, 1993.

	Date											
	19	20	21	22	23	24	25	26	27	28	29	Total
No. males	2	0	1	2	1	4	6	2	2	2	0	22
No. females	0	0	0	0	0	0	2	0	0	0	0	2

Table 6. Density of residual sockeye trapped in 1993 at Redfish Lake.

Length (mm)	Weight (g)	Fish #	Sex	Destiny[a]
222		1	M	Milt taken, released 10/25
214		2	M	Milt taken, released 10/25
215		3	M	Milt taken, released 10/25
225		4	M	Milt taken, released 10/25
214		5	M	Milt taken, released 10/25
226		6	M	Milt taken, released 10/25
215		7	M	Milt taken, released 10/25
225		8	M	Milt taken, released 10/25
208	89.4	1	FM	9.5 g eggs fert w/resm 1&2
208	103.5	1	M	spawner
217	110.9	2	M	spawner
215	95.9	2	FM	11.7 g eggs, fert w/resm 3&4
220	104.8	3	M	spawner
207	93.5	4	M	spawner
210	99.0	9	M	Milt taken, sampled 11/1
215	97.5	10	M	Milt taken, sampled 11/1
208	93.6	11	M	Milt taken, sampled 11/1
225	115.2	12	M	Milt taken, sampled 11/1

[a]Fourteen male sockeye were released unharmed.

Table 7. Mating crosses made with anadromous, captive broodstock, and residual adults of Redfish Lake stocking during the period of October 1993 through April 1994. Number of females listed first, followed by the number of males used by genetic origin.

Brood Year	Males Spawned	Females Spawned				
		ANAD	OM91	OM92	BY91	RES
BY 93 (October-November)						
	ANAD	2 x 7[a]	1 4 x 6			
	OM91		4 x 2			
	OM92					
	BY91		8 x 3		1 x 2	
	RES					2 x 4
BY 93.5 (December-April)						
	ANAD					
	OM91		2 x 2			
	OM92					
	BY91				5 x 8	
	RES					

[a] Cryopreserved sperm used for one lot per female.
[b] Cryopreserved sperm used since no fresh sperm was available.

A "safety net" broodstock was established from these matings with 35 eggs per surviving egg lot going to BBC and 25 eggs per lot to EAG. The remainder are being grown at EAG for release into Redfish Lake.

Mating of Residuals

Two female residuals were mated with four male residuals which were trapped from Redfish Lake (Table 6). The resulting eyed eggs were divided between BBC and EAG for broodstock purposes. The goal is to produce a small number of F, progeny to evaluate whether this stock has potential use for recovery.

Brood Year "93.5" Matinas (January to April 1994)

A total of 5 females in the captive broodstocks reared at EAG matured during this unusual time period (Appendix B). Males also matured at this time and were used whenever possible for fresh sperm. At times, cryopreserved sperm of residuals was used for fertilization. Also, gonadotropin releasing hormone (GnRH) pellets were implanted into females and males to synchronize maturation. Surviving progeny of these matings will be released into Redfish Lake when they attain a size which approaches that of a presmolt.

Because the fertilization rate of eggs spawned in the fall of 1993 was poor, an investigation was conducted on problems associated with the maturation of these fish (Table 8).

Drs. Carl Schreck (Oregon State University).and Penny Swanson (NMFS) provided input for the samples to be taken. Blood was drawn from the caudal peduncle into a heparinized 3 ml syringe and centrifuged. The plasma was then drawn off and frozen at -80°C. Two males and two females were given a GnRH implant in the dorsal musculature on January 31, 1994. The male which provided milt on January 31 was not induced, nor was the female. These eggs appeared to be of poor quality due to some "moon-eyes" in those expressed. The milt sample obtained from male #4F was examined for motility in well water and Thorgaard's activator, and a sample was centrifuged for spermatocrit. One sample of this milt was fixed in Davidson's fixative (AFA) and a second was frozen at -80°C, each labeled by PIT-tag numbers.

RELEASE OF ADULT OM91 TO REDFISH LAKE

Twenty-four 1991 Redfish outmigrant broodstock adults were selected for return to Redfish Lake based on fork length and maturity determined by ultrasound on August 8, 1993 (see ultrasound section of Captive Broodstock Research, this report). The release group included 12 of each sex. Females were given a second PIT-tag on the leading edge of the

Table 8. Documentation of female maturation condition for broodstocks of Redfish Lake sockeye salmon, Eagle Hatchery, 1994. All samples referenced by bar code labeled with date, broodstock type, and PIT tag number.

Date	Type	PIT tag number	Diet Reg/Sp	FL (mm)/ wt (g)	Ova Weight	Dia (mm)/ wt (#/g)	Samples (Eggs) Plsm)	% Eye
2/I /94	OM91	7F7F507E7E	Reg	505/I217	55.7	3.88119.2	B7-10/85.6	0.0
2/3/95	OM91	7F7F525D55	Reg	550/1736	49.0	4.75/12/2	B12,13/BII	0.0
2/3/95	OM91	7F7F582046	Reg	580/I790	183.4	5.0110.8	B15,19/614	0.5

dorsal fin, and all fish were implanted with an ultrasonic tag inserted into the stomach. These were observed for at least two days for tag retention. Releases were made on August 10, 11, and 12, 1993. Each load was tempered from 10.4°C to 16.6°C over a 2.25-h period using Redfish Lake water. For each release, fish handled the transport, acclimation, and release well. Tracking of the released group will begin during the third week of August (Kline 1994).

The following summarizes pertinent data:

Release Date	ABD PIT	DOR PIT	Sex	Fork Lenath	Sonic Tan	Comments
8/10,11,&12	7F7F51040F		M	480	2273	
	7F7F531626	7F78094829	F	485	2327	
	7F7F576238	7F7BOAOD59	F	530	2246	
	7F7F534F6E		M	508	2264	
	7F7F582835	7F78097453	F	475	2255	
	7F7F583025		M	492	366	
		7F78077209	F	520	357	
	7F7F525E06		M	465	348	CATARACT
	7F7F525D39		M	505	294	
	7F7F531430	7F7B077F35	F	552	339	
	7F7F525E05	7F78077A6B	F	528	285	
	7F7F53073D		M	480	276	
	7F7F552B4E	7F7BOF260C	F	525	249	
	7F7F51041B	7F7B08471C	F	518	258	
	7F7F535363		M	495	267	
	7F7F414210	7F7D451069	F	518	456	
	7F7F581129		M	488	447	
	7F7F574F30	7F78076341	F	544	465	
	7F7F525A46		M	492	375	
	7F7F55281A		M	480	384	
	7F7F552F4F	7F78081614	F	492	2534	
	7F7F551511		M	440	2525	
	7F7F582267	7F7B08467E	F	490	2453	
	7F7F551868		M	465	2444	

CAPTIVE BROODSTOCK RESEARCH

The result of the first spawning of the Redfish Lake sockeye captive broodstock in 1993 was disappointing. The overall fertilization rate was 30% using milt from feral anadromous males crossed with eggs of females reared from smolts to maturity. The fertilization rates were even lower when milt was used from males reared in captivity. These males were both outmigrant and BY91 sockeye origin. The spawning protocols used were

those developed for sockeye by the Alaska Department of Fish and Game (McDaniel et al. 1994) and have been used successfully by IDFG with anadromous adults. One aspect of these protocols (i.e., iodophore disinfection) was tested in 1993 and found not to be at fault.

The National Marine Fisheries Service has been rearing a surrogate sockeye broodstock at Manchester, Washington to investigate broodstock culture techniques prior to spawning of the portion of the Redfish Lake sockeye broodstock being reared there. Some adults were spawned from the surrogate stock in 1993 yielding an average fertilization rate of 36%. These low fertilization rates will hamper recovery efforts of the ESA listed stock unless dramatically improved.

There was a meeting of the Broodstock Nutrition Work Group in February 1993 to make suggestions on the conduct of the sockeye captive broodstock program. This group consists of culturists and nutritionists from around the northwest, and the purpose is to discuss ways to improve female quality in captive broodstock. Several members of this group have given suggestions to diet modifications that improve the quality and subsequent fertilization rate of gametes originating from captive adults.

Also, southwest Idaho has been determined to be extremely deficient in the element selenium, and the livestock industry has demonstrated that supplementation is desirable (Lane, personal communication, 1994). The diet which has been fed to the sockeye salmon in the captive broodstock program has a selenium level of 0.1 mg/kg, and the manufacturer (Bioproducts, Inc.) has stated that a higher level would be desirable to optimize reproductive success.

MODIFIED DIET

The modifications are based on some experimentation and some intuitive information and are as follows:

Diet base is to be Biodiet Brood Diet modified with replacement of 50% of the fish meal with Antarctic krill meal, and the addition of selenium at 2.5 mg/kg diet as All-tech, Inc. yeast formulation, Vitamin E at 30 iu/kg, and Vitamin C at 2600 iu/kg.

TEST FACILITIES

Eagle Fish Hatchery

Water used to rear sockeye broodstock originates from up to seven artesian wells and is pumped through degassing towers to strip excess dissolved gas. Eagle Hatchery was converted to culture the ESA listed sockeye beginning in 1991, and had served as a production station for resident species since 1938. Water temperature varies only slightly

from 11.5°C to 13.3°C during the year. Containers for rearing include 12, 3-m circular tanks, 4,4-m semisquare tanks, and up to 20, 2-m tanks. The total water used at this station is 0.2 m^3/sec. Effluent from these tanks enters a series of two settling ponds on the premises which have a retention time of about 2.2 h and is discharged into a creek with a flow of approximately 0.5 m^3/sec. This creek flows through agriculture land for a distance of three miles before. entering the south channel of the Boise River.

Inventories and the proportion of the test fish for the proposed trial are as follows:

Redfish Lake sockeye outmigrant 1991: 186 fish at 1600 g with 75% fed the test diet;

Redfish Lake sockeye outmigrant 1992: 70 fish at 800 g with 100% fed the test diet (one tank used);

Redfish Lake sockeye anadromous BY1991: 860 fish at 950 g with 75% fed the test diet.

Fish are fed four times a day for optimal feed consumption and at a rate which is about 70% of the manufacturer's recommended level. The circular configuration of tanks provides optimal water use and tanks are self-cleaning. Tanks are swept weekly. The goal of the growth program is to produce maturity in the fourth and fifth year for outmigrant groups and in the third and fourth year for fish reared from eggs. The culture of sockeye broodstocks is in its infancy, so these growth programs are based on ongoing performance monitoring of the stocks. The proposed level of selenium added to the diet in the IDFG test will be equal to a total of 7.35 g selenium fed over the period of April to September 1994. This period was selected because fish nutritionists feel that this would be a sufficient time to provide mobilization of the altered dietary selenium and other components to effect the desired elevation of fertilization rate.

EVALUATION

Parameters which will be monitored during the test period include levels of selenium, vitamin E, and vitamin C in the feed; in plasma, gonads, and muscle from mortalities; fatty acid profiles in feeds and fat reserves in mortalities; and gill structure and ceroid in liver if the suspected problem is nutritional gill disease. Parameters which will be investigated in treated and control groups during spawning include fecundity, fertilization rate within the spawning matrix, egg diameter and weight, sperm motility, genetics, and pathology.

Periodic nonlethal blood samples may be taken to track plasma levels of sex hormones in test and control groups and may also be used for the added diet components. Laboratories which will do the testing have not as yet been identified.

DATA ANALYSIS

Data will be analyzed using Systat. The General Linear Models procedure will be used to test for main effects and interactions.

ULTRASOUND EXAMINATION OF 1991 OUTMIGRANTS FOR MATURITY

The 1991 outmigrants from Redfish Lake and Alturas Lake were examined for sexual maturity with ultrasound to select fish with a high probability for spawning. The goal was to obtain a group from each lake for release for volitional spawning.

The procedure was as follows:

1. Remove individual fish from the vat at SFH.
2. Anesthetize fish in MS-222.
3. Read the PIT-tag and record on the ultrasound tape.
4. Examine the body cavity for gonad type and development.
5. If mature, measure fork length.
6. If female, install a second PIT-tag on the dorsal fin.
7. Insert a sonic or radio tag into the stomach and record the frequency of the tag and the PIT-tag(s) number.
8. Return the fish to a separate holding area.

Results

The degree of maturity was more advanced for the Alturas group than for the Redfish group. A total of 24 Redfish and 20 Alturas outmigrants were showing enough maturity for release with a high probability for spawning. The Alturas group was expected to be more advanced because the expected spawning time is early September, while that of the Redfish group is mid-October. We tried to select fish which had a representative variation in apparent degree of development of gonads rather than selecting only those with advanced gonad development. This was to attempt to get a range in spawning time if that was the cause of the gradation in development. All fish took the handling well and there was no regurgitation of tags 48 hours after they were implanted in the Redfish Lake group. Radio tags will be implanted in the fish for release into Alturas Lake when the tags arrive. Tracking and performance of tagged fish is reported by Kline (1994).

AKNOWLEDGEMENTS

The Redfish Lake Captive Broodstock program has been operational for three years. As in past years, many experts have lent their time and efforts to these fish and provided many suggestions which have contributed to progress made to date. Their efforts have been greatly appreciated.

Funding has been provided by the Bonnevilte Power Administration through Dr. Jeffrey Gislason, Contracting Officer, who has arranged funding for a major remodeling of the Eagle Fish Hatchery water supply to provide chilled water for adult holding prior to final maturity. This has been anxiously awaited for several years and soon will become a reality.

Fish culturists from several IDFG hatcheries have provided support for this program by assisting in smolt trap and adult weir monitoring and spawning. The staff of Sawtooth Hatchery have provided expert assistance consistant with the sensitive nature of this project. The staff of the Eagle Fish Health Laboratory has processed an exorbant number of sockeye cases and the results of these have assisted in prompt attention to the health need of these fish. Training in application of PIT tags and the associated databases was provided to our staff by members of Russ Kiefer's Idaho Parr and Smolt Monitoring project. This has allowed our staff to become more independent of others in the maintenance of this important function.

Special thanks to Paul Kline for critically reviewing this manuscript and all his help during the year with fish culture activities.

Return of eight anadromous adult sockeye enabled us to greatly expand our genetic base. These fish have provided hope that these broodstocks can still be maintained to preserve the unique characters of this stock.

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A P P E N D I C E S

Appendix A. Final fertilization rates by family for Redfish Lake sockeye outmigrant and anadromous adults.

Date	Group	FL	PIT TAG	Groups by male							Fecundity
10/7	l&2	528	7F7F574E5C	1+2,	3+4,	5+6					1679
			GREEN EGGS	560	560	560					
			% FERT.	51	0	0					17
			EYED EGGS	285	0	0					285
			EAG	250							
			BBC	35							
10/8	A	505	7F7B09382A	1	2	3	4	5	6	LL	3160
			GREEN EGGS	500	500	500	500	500	500	150	
			% FERT.	72	9	74	81	25	70	0	52
			EYED EGGS	351	45	371	406	125	351	0	1649
			ID FM	A	A	A	A	A	A	A	
			ID M	A	A	A	A	A	A	A	
			DESTINY EAG	247	47	257	292	63	237	0	1143
			BBC	114	0	114	114	62	114	0	518
	B	505	7F7BOA6B5B	1	2	3	4	5	6	LL	3159
			GREEN EGGS	500	500	500	500	500	500	150	
			% FERT.	90	23	93	93	20	90	0	65
			EYED EGGS	450	117	466	463	102	452	0	2050
			ID FM	A	A	A	A	A	A		
			ID M	A	A	A	A	A	A		
			DESTINY EAG	336	0	352	349	0	338	0	1359
			BBC	114	117	114	114	102	114	0	675
10/9	l&2	522	7F7F7A531C	1	2	5				2600	
			GREEN EGGS	870	870	870					
			% FERT.	0	41	30				24	
			EYED EGGS	0	360	260				620	
			ID FM								
			ID M	A	A	A					
			DESTINY EAG	0	175	125					
			SFH		150	100					
			BBC		35	35					
10/14	3&5	468	7F7F413176	3	4	5				1389	
			GREEN EGGS	460	460	460					
			% FERT.	73	26	54				51	
			EYED EGGS	325	120	250				705	
			ID FM								
			ID M	A	A	A					
			DESTINY EAG	150	60	125					
			SFH	150	60	125					
			BBC	35							
10/17	3&5	465	7F7F582C46	5	1	8				1567	
			GREEN EGGS	520	520	520					
			% FERT.	55	48	0				34	
			EYED EGGS	285	250	0				535	
			ID FM								
			ID M	A	A	OM					
			DESTINY EAG	125	125						
			SFH	125	125						
			BBC	35							

l&2	585 7F7F53605B		7	1	8	2953
	GREE N EGGS		990	990	990	
	% FERT.		0	29	0	10
	EYE D EGGS		0	285	0	285
	ID FM					
	ID M		A	A	OM	
	DESTINY	EAG		250		
		SFH				
		BBC		35		
l&2	529 7F7DOE014C		1	4	6	2207
	GREE N EGGS		735	735	735	
	% FERT.		41	59	21	40
	EYE D EGGS		300	435	155	890
	ID FM					
	ID M		A	A	A	
	DESTINY	EAG	150	200	60	
		SFH	150	200	60	
		BBC		35	35	
l&2	459 7F7F513565		2	3	6	1495
	GREE N EGGS		500	500	500	
	% FERT.		60	77	50	63
	EYE D EGGS		300	385	250	935
	ID FM					
	ID M		A	A	A	
	DESTINY	EAG	150	125	125	
		SFH	150	125	125	
		BBC		35		
l&2	551 7F7F535FOF		6	7	2	1655
	GREE N EGGS		550	550	550	
	% FERT.		11	0	28	13
	EYE D EGGS		60	0	155	215
	ID FM					
	ID M		A	A	A	
	DESTINY	EAG	25		60	
		SFH			60	
		BBC	35		35	
l&2	520 7F7F527154		2	5	1	4034
	GREE N EGGS		1345	1345	1345	
	% FERT.		47	29	45	40
	EYE D EGGS		635	385	600	1620
	ID FM					
	ID M		A	A	A	
	DESTINY	EAG	300	150	300	
		SFH	300	150	300	
		BBC	35	35		
l&2	514 7F7DOB3119		6	7	2	2831
	GREE N EGGS		940	940	940	
	% FERT.		62	15	53	43
	EYE D EGGS		580	137	5.00	1217
	ID FM					
	ID M		A	A	A	
	DESTINY	EAG	270	102	250	
		SFH	275		250	
		BBC	35	35		

	l&2	498	7F7DOD604E		2	3	5		2042
		GREE N EGGS			680	680	680		
		0 FERT.			9	49	50		36
		EYE D EGGS			60	335	340		735
		ID FM							
		ID M			A	A	A		
		DESTINY	EAG			150	170		
			SFH		60	150	170		
			BBC			35			
	4	467	7F7F50721E		1	2	4		1337
		GREE N EGGS			445	445	445		
		% FERT.			19	16	39		25
		EYE D EGGS			85	70	175		330
		ID FM							
		ID M			A	A	A		
		DESTINY	EAG		50	70	140		
			SFH						
			BBC		35		35		
	4	508	7F7F414653		1	5	6		2171
		GREE N EGGS			720	720	720		
		% FERT.			41	46	47		44
		EYE D EGGS			295	330	335		960
		ID FM							
		ID M			A	A	A		
		DESTINY	EAG		130	165	150		
			SFH		108	149	138		
			BBC		35		35		
10/19	4	515	7F7F507107		7	8	9	1	1435
		GREE N EGGS			470	470	470	106	
		% FERT.			0	29	5	50	
		EYE D EGGS			0	135	35	53	
		ID FM							
		ID M							
		DESTINY	EAG						
			SFH			100		53	
			BBC			35	35		
10/21	4	526	7F7F552E7A		5	7	9		2351
		GREE N EGGS			695	728	620		
		% FERT.			1	0	0		
		EYE D EGGS			10	0	0		
		ID FM							
		ID M							
		DESTINY	EAG						
			SFH		10				
			BBC						
11/8	3&5	480	7F7F553469		2	5	10		1210
		GREE N EGGS			403	403	403		
		% FERT.			0	0	0		
		EYE D EGGS			0	0	0		
		ID FM							
		ID M							
		DESTINY							

l0/28 RESIDUA L SOCKEYE FEMALE A CROSSED WITH POOLED MILT l&2

GREENN EGGS	160
% FERT	86
EYEDD EGGS	138
DESTINY	
EAG	68
BBC	68

RESIDUA L SOCKEYE FEMALE B CROSSED WITH POOLED MILT 3&4

GREEN EGGS	157
% FERT	98
EYED EGGS	154
DESTINY	
EAG	77
BBC	77

Appendix B. Fertilization rate as percent eye for late spawning pairs from captive sockeye BY91 and OM91 females crossed with fresh and cryopreserved sperm.

Spawn Date	Group	FM PIT (last 3#)		Males by PIT #		(Fertilization medium)
12/29	BY91	51D		F68+DlF	30E+BO B	62A+463 (HW)
				TOTAL SWIM-UP WAS 5.6%		
2/l	OM91	E7E		54F	053	(Hw
			EG G NO.	577	461	
			EYED NO.	0	0	
			% EYED	0	0	
2/3	OM91	D55		54F	053	(HW)
			EGG NO.	328	328	
			EYEG NO.	0	1	
			% EYED	0	0	
		046		54F	053	(HW)
			EGG NO.	1192	1192	
			EYED NO.	0	55	
			% EYED	0	4.6	

Appendix C. Permit modifications obtained.

The following modifications to permit # 795 were obtained during the reporting period.

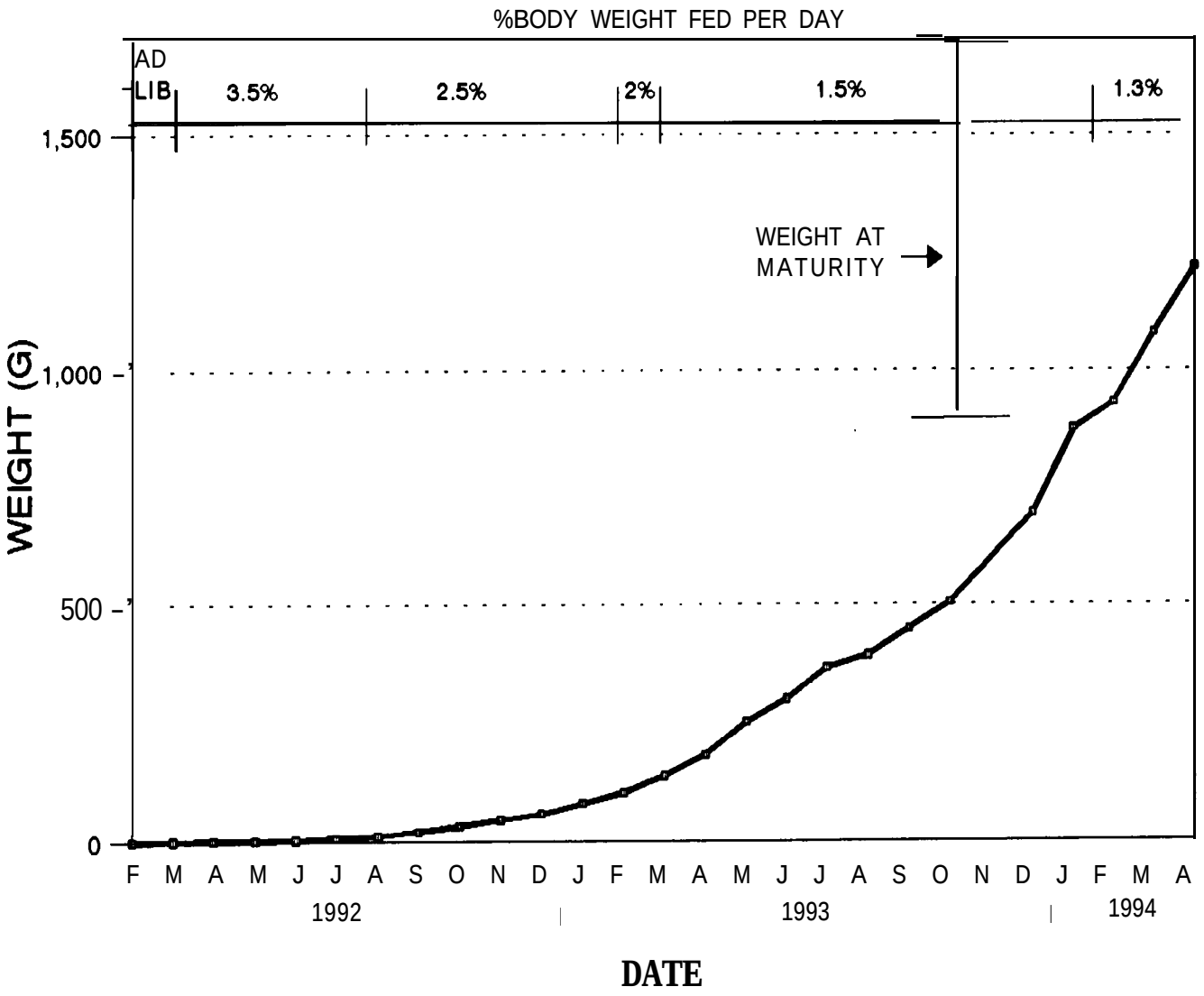
1. Modification 1 to Permit #795 issued August 3, 1993 to allow radio tagging of adults from captive broodstock program and their release into Redfish Lake.

2. Modification 2 to Permit #795 issued October 8, 1993 to allow collection or residual sockeye salmon and release of progeny of 1991 outmigrants into Redfish lake in 1994.

APPENDIX D.

Growth rates for broodstocks.

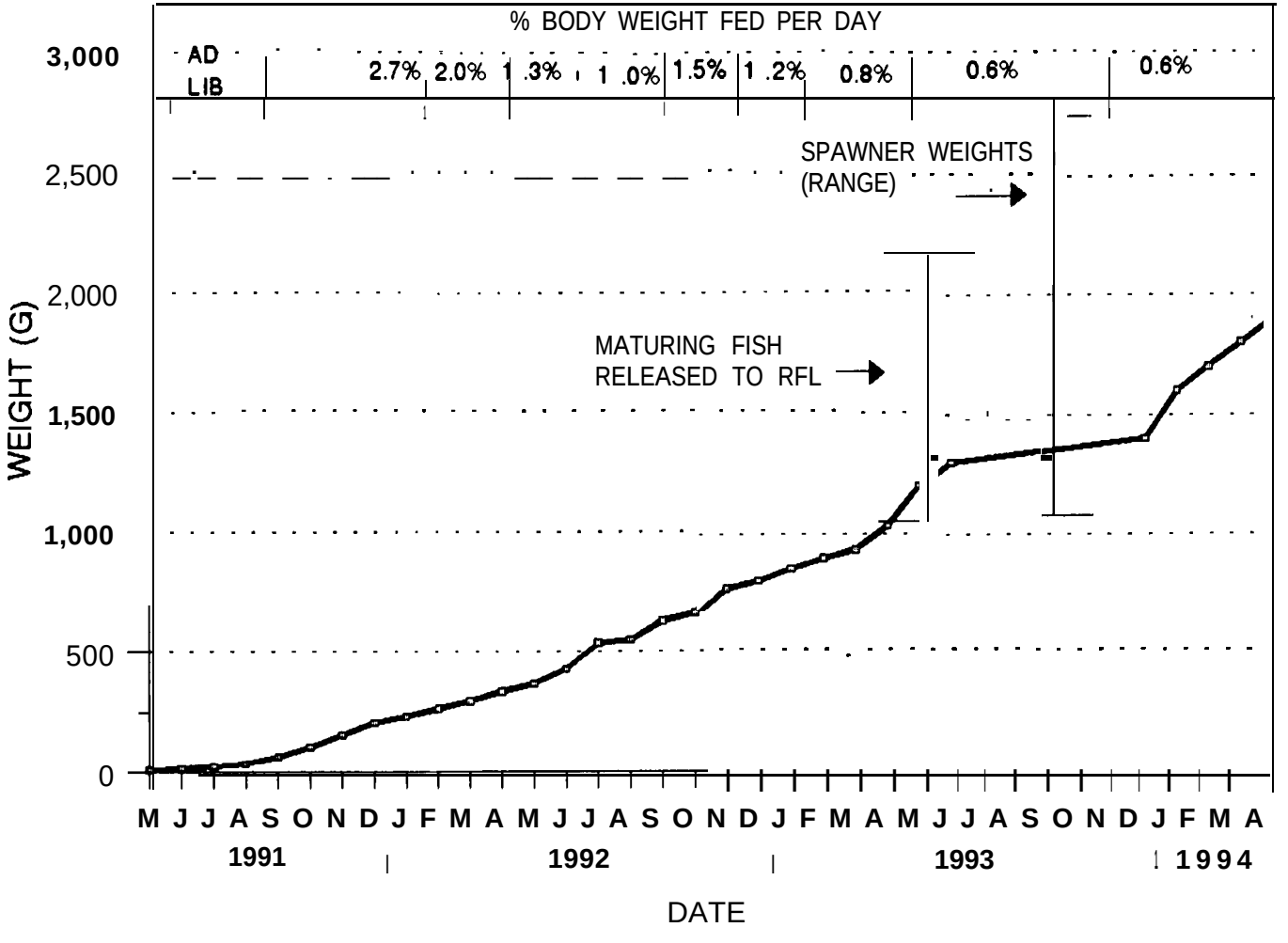
RFL BROOD YEAR 1991 GROWTH, FEED RATE, AND SIZE AT MATURITY I/92-4/94



Growth feed rate,and size at maturity of BY91

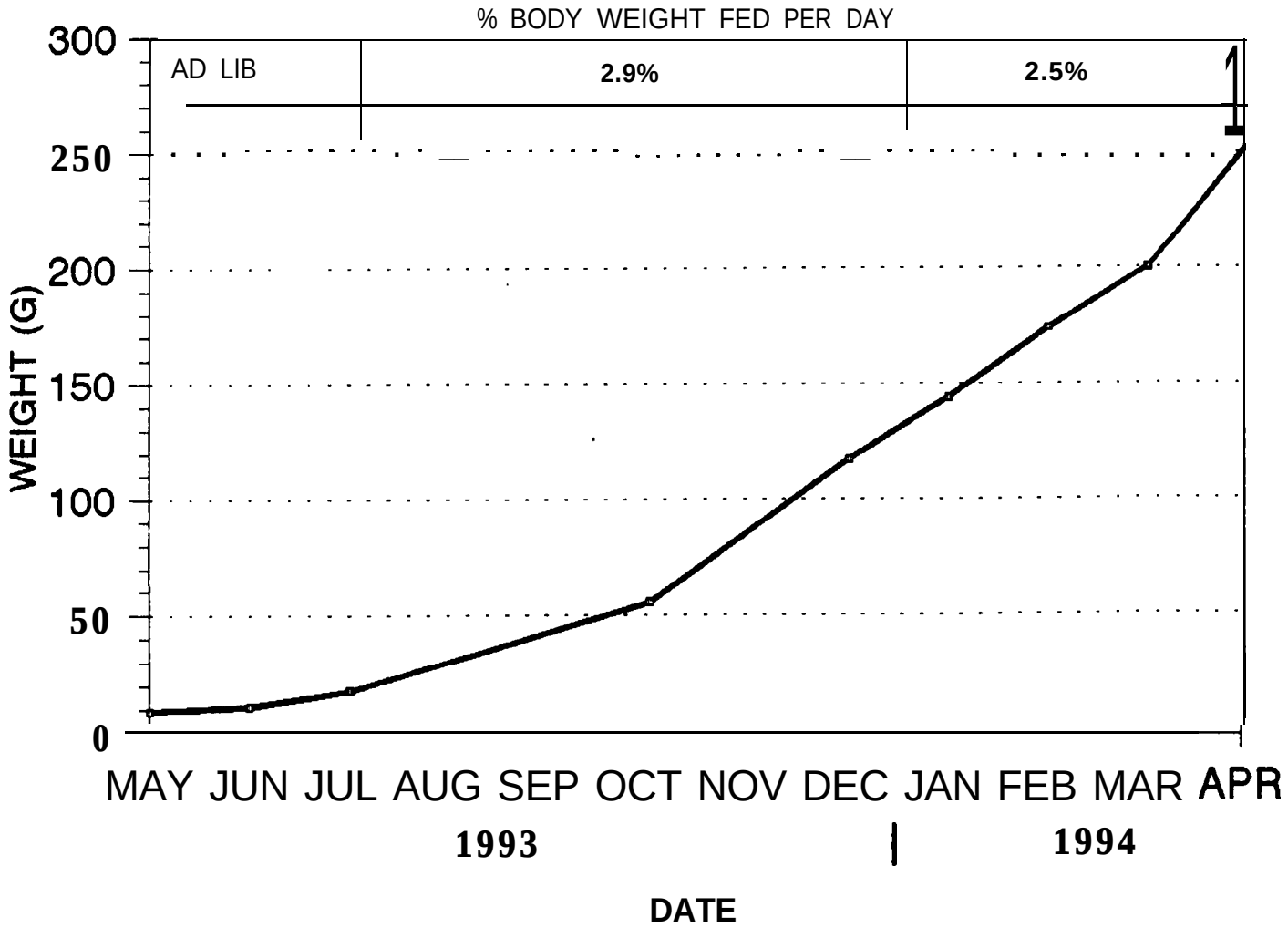
RFL OUTMIGRANT 91 GROWTH, FEED RATE, AND SIZE AT MATURITY

5/91-4/94



Growth, fed rate, and size at maturity for OM91

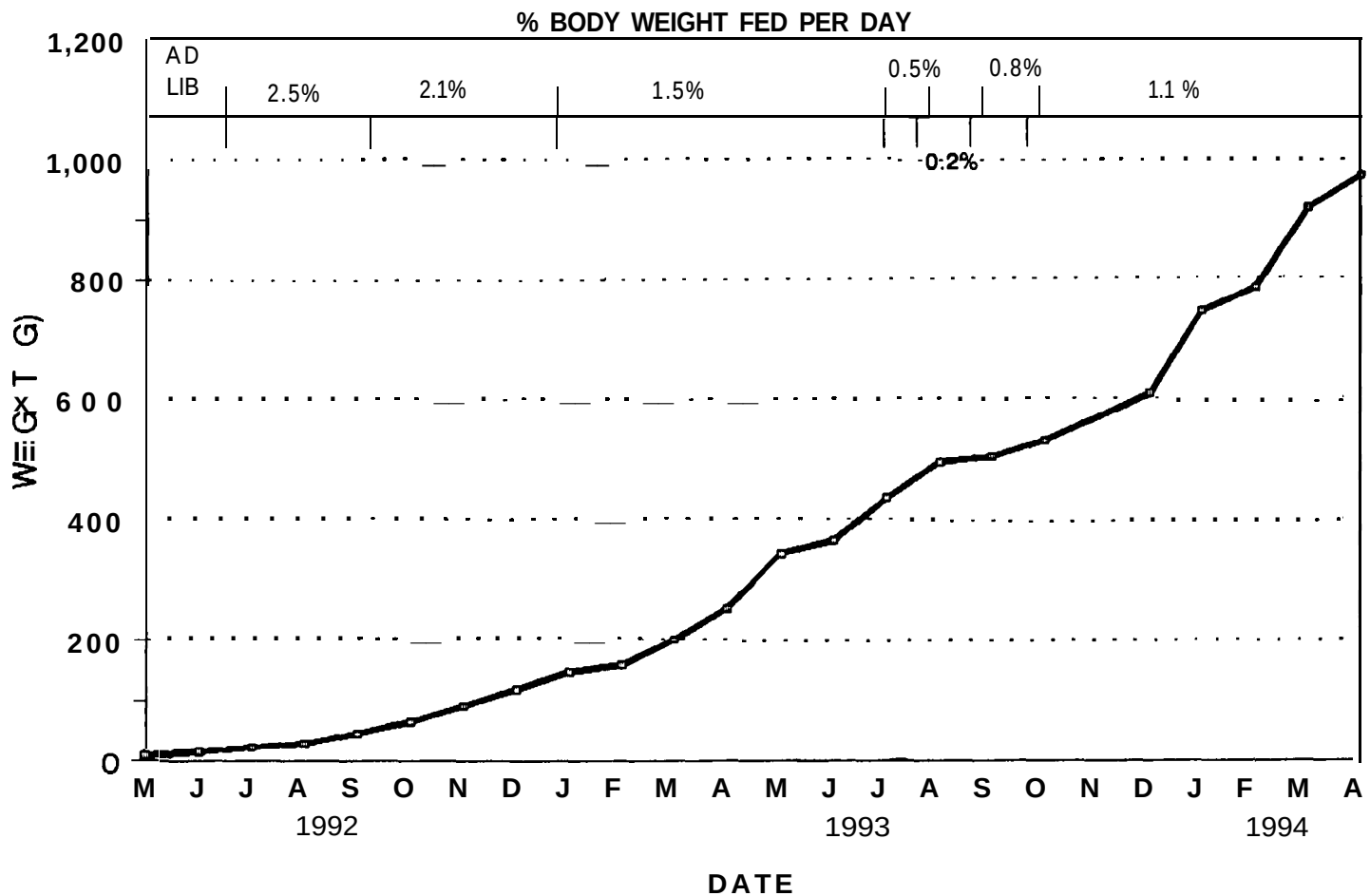
RFL OUTMIGRANT 93 GROWTH AND FEED RATE
5/93-4/94



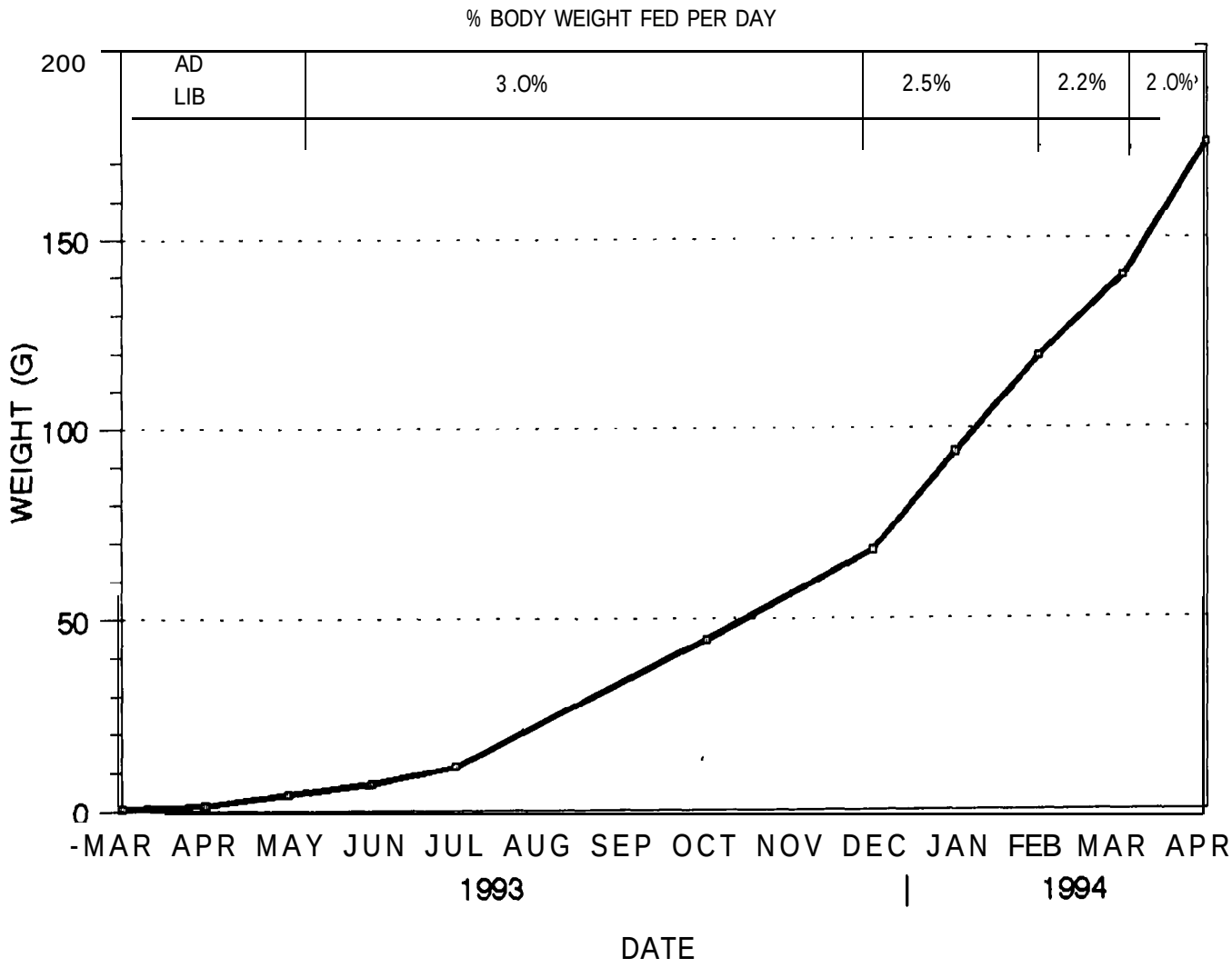
Growth and feed rate for OM93.

RFL OUTMIGRANT 92 GROWTH AND FEED RATE 5/92- 4/94

Growth and feed rate for OM92.



RFL RESIDUAL BROOD YEAR 1992 GROWTH AND FEED RATE 3/93-4/94



Growth and feed rate for REBY92.

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